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The Effect of Pasak Bumi (*Eurycoma longifolia*, Jack) Roots Ethanol Extract against Hematology Profile of Healthy Volunteers

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ABSTRACT

The roots of *Eurycoma longifolia*, Jack has a lot of beneficial activity on human health. The use of *Eurycoma longifolia*, Jack roots as a traditional medicine should be made in the form of a dosage that is more effective, safe, and had no side effects, especially on hematology. Easy preparations are a capsule. This study aimed at identifying the effect of ethanol extract capsule of *Eurycoma longifolia*, Jack roots on hematology of healthy volunteers. Healthy volunteers were assigned in the study, each of which was 10 male and 10 female who met the inclusion criteria. Both groups were given ethanol extract capsule of *Eurycoma longifolia*, Jack roots for 14 days, with the dose of 300 mg extract once a day, after meal, at night. Hematology examination was performed on day 0; 14 and 42. Results were compared statistically using Repeated ANOVA and Friedman tests. The result of the study indicated that the average value of hematology at the examination day of 0; 14; and 42, there were significant differences in MCV and leukocyte parameters in healthy male volunteers ($p < 0.05$). MCH, LED and monocyte parameters in healthy female volunteers had a significant difference of ($p < 0.05$). However, the parameter values were still within the normal range. Monitoring results showed that some of the side effects were frequent urinary, dizziness, nausea, sweating, insomnia, and increasing appetite on healthy male volunteers. Meanwhile, on healthy female volunteers, there were frequent urinary, insomnia, constipation. Having ethanol extract capsule of the *Eurycoma longifolia*, Jack roots did not affect the hematology parameters of healthy male and female volunteers, but it caused some side effects.

Keywords: *Eurycoma longifolia*, capsule, safety, hematholog, healthy volunteer

INTRODUCTION

Pasak bumi (*Eurycoma longifolia*) is one of the many herbs in Southeast Asia, including Indonesia, Malaysia, Thailand, Laos, Cambodia and Vietnam (Hassanah *et al.*, 2006). *Pasak Bumi* is widely used as antioxidant, malaria drug, health supplement, anticancer, and aphrodisiac (Rehman *et al.*, 2016). Almost all parts of this plant can function as a medicine. Compounds contained in *Pasak Bumi*, among others, are cuasinoids (Miyake *et al.*, 2009), triterpenes (*tirucallane*), *squalene* derivatives, eurycolactone, laurycolactone, eurycomalactone (Bhat *et al.*, 2010), 9-methoxisanthin-6-on alkaloids (Rosli *et al.*, 2009), and flavonoids (Nurani, 2008).

Research on pharmacological root activity has been widely carried out, pre-clinical trials have reported that ethanol extracts of root of *Pasak Bumi* given to SD female rats for 14 days has been shown to give effect as an immunostimulator with a mechanism to significantly increase lymphocyte proliferation activity compared to the control group (Falah *et al.*, 2011). The research conducted at the Bogor Institute of Agriculture shows that the

roots of *Pasak Bumi* have the property to protect the liver from damage (Panjaitan, 2008).

Meanwhile, the research conducted by Al-Salahi *et al.*, (2012) showed that the administration of methanol extract of root of *Pasak Bumi* for 16 days to male rats did not affect the hematological profile (hemoglobin, erythrocytes, hematocrit, leukocytes, platelet count, cell volume, and corpuscular cells).

The 90 days acute, subacute, and subchronic toxicity tests of the water of the extracted *Pasak Bumi* roots did not show significant changes in hematologic parameters, namely erythrocytes, leukocytes, hemoglobin, and hematocrit in either male or female rats compared with the control group (Choudhary *et al.*, 2012). Shuid *et al.*, (2011) reported that the subacute toxicity test of *Pasak Bumi* root water extract that had been carried out had no significant differences in the hematological parameters (erythrocytes, hemoglobin, MCV, MCHC, platelets, white blood cell count, lymphocytes, monocytes, neutrophils and eosinophils) between the control group and the treatment group, as well as the morphology of normal red blood cells in all test animals. Clinical trials conducted by Hayati (2013) showed the results of tolerability tests in humans indicated

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that the administration of standardized *Pasak Bumi* root water extracts at a dose of 1.2 grams twice daily for 14 days was safe and did not cause harmful side effects. The pilot study conducted by Henkel *et al.*, (2013) showed that the administration of standardized *Pasak Bumi* water extract at a dose of 200 mg twice daily for 5 weeks in male and female subjects as a health supplement did not have side effects on the hematological parameters. However, Li *et al.*, (2013) stated that long-term consumption of *Pasak Bumi* roots can cause sleeplessness, redness of the face, pressure on the testes, and hyper-aggressive. Nonetheless, there is no scientific evidence to support these side effects.

The results of previous studies conducted on the use of extracts of the *Pasak Bumi* root water extract as traditional medicine did not show any harmful side effects. Today, the traditional medicine industry is starting to develop medicinal plants not only in powder form but also in extract form to ensure the preparations contain active ingredients with effective doses for therapy. The drug industry usually produces such extracts using harmless solvents, such as ethanol (Serlahwaty *et al.*, 2008). The problem of extracting natural ingredients tends to be a bad taste and distinctive odor. Currently, preparations that are easily made and able to smother the bitter taste of the roots of *Pasak Bumi* and are more storage-practical, as well as to cover the shortage of natural ingredients, are in capsule form (Syamsuni, 2007).

Based on the description above, further research is conducted on the effect of ethanol extracts of *Pasak Bumi* in the form of capsules on the hematological picture of healthy volunteers is aimed to determine its effect on hematological value, whereas, the phase I clinical trials of hematological examination is used to observe the safety and toxic effects of drugs on healthy subjects (Santoso, 2006). A hematological examination can indirectly monitor the state of the body (Ifeyani *et al.*, 2014). Hematologic tests include hemoglobin, erythrocytes, hematocrit, leukocytes, platelets, leukocyte counts (lymphocytes, monocytes, neutrophils, eosinophils, basophils), Erythrocyte Index (MCV, MCHC), Red Cell Distribution Width (RDW), and the rate of the sedimentation of blood (LED).

METHODOLOGY

Subject Selection Criteria

The selection of the volunteers in this study had met the inclusion and exclusion criteria. The inclusion criteria were men and women aged 18-42 year-olds willing to be subjects (filled in informed consent), healthy as evidenced by health

certificate from a doctor who has a license to practice, do not smoke, not taking propranolol, hypoglycemic drugs, vitamins, or health supplements (Salman *et al.*, 2010). The exclusion criteria were subjects who dropped out while the study was underway, BMI more than 30 kg/m², and pregnant and nursing women.

Method of Clinical Research

The hematological parameter values of the healthy volunteers were measured before day 0, after day 14 of taking capsules of ethanol root extracts of *Pasak Bumi*, and after 28 days of not consuming capsules of ethanol extracts of root *Pasak Bumi* (day 42). The monitoring based on clinical and psychological symptoms was performed throughout. Capsules were given at a dose of 300 mg extract, taken once a day after meals or at night before going to sleep for 14 days (Kanokkangsada, 2016).

Hematologic parameters measured in this study include hemoglobin, erythrocytes, hematocrit, leukocytes, platelets, leukocyte type counts (lymphocytes, monocytes, neutrophils, eosinophils, basophils), Red Cell Distribution Width (RDW), Erythrocyte Index (MCV, MCH, MCHC), and sedimentation rate (LED). The hematological examination was carried out at the Parahita Diagnostic Center.

RESULTS AND DISCUSSION

This study has received a statement of ethical conduct from the Muhammadiyah University Ethics Commission, Yogyakarta.

Overview of Hematology Profiles of Healthy Volunteers after Giving Capsules of Root Extract of *Pasak Bumi*

The result of the statistical analysis (Table I) of hematology examination on day 0, day 14, and day 42 showed no significant difference in healthy male volunteers ($p > 0.05$) in hematologic parameters, namely hemoglobin, erythrocytes, hematocrit, platelets, count type of leukocytes (lymphocytes, monocytes, neutrophils, eosinophils), erythrocyte index (MCH, MCHC), Red Cell Distribution Width (RDW), and sedimentation rate (LED), but showed no significant difference ($p < 0.05$) in the parameters of leukocytes and MCV. There was no significant difference in the parameters of leukocytes ($p = 0.008$) and MCV ($p = 0.047$) seen in the 14th day and 42nd-day examination, but the values of the two parameters were still in the normal range.

The tests on day 0 and day 14 to see the effect of ethanol extract capsules of roots of *Pasak Bumi* for 14 days showed no significant difference

Table I. Mean Values of Hematology Day 0, Day 14 and Day 42 in Healthy Male Volunteers (n=10)

Hematology Parameters	Mean Values of Hematology of Healthy Male Volunteers (Mean±SD)			P value	Normal Value
	Day 0	Da 14	Day 42		
Hemoglobin (g/dL)	15,58±0,82	15,46±0,86	15,54±0,84	0,475	13,2-17,3
Erythrocytes (million/uL)	5,32±0,37	5,30±0,35	5,33±0,34	0,590	4,4-5,9
Hematocrit (%)	43,55±2,47	43,53±2,09	43,61±1,95	0,910	40-50
Platelets (10 ⁴ /uL)	26,17±2,49	25,77±3,45	26,15±3,07	0,788	15.0-44.0
MCV (fl)	82,06±3,29	82,24±3,27	82,01±3,22	0,038*	80-100
MCH (pg)	29,25±1,06	29,21±1,21	29,20±1,06	0,885	26-34
MCHC (g/dL)	35,66±0,92	35,51±0,84	35,63±0,83	0,387	32-36
RDW (%)	13,00±0,61	13,05±0,54	12,94±0,50	0,178	11.5-14.5
LED (mm)	11,30±13,48	9,90±8,27	9,00±13,03	0,245	0-10
Leukocytes (10 ² /uL)	64,73±10,29	62,18±13,27	69,70±12,36	0,033*	38.0-106.0
Lymphocytes (%)	40,20±4,13	41,50±6,33	37,90±5,88	0,295	25-40
Monocytes (%)	6,90±1,52	7,90±2,13	8,10±0,88	0,145	2-8
Neutrophils (%)	49,10±4,48	47,30±7,76	50,50±6,02	0,502	50-70
Eosinophils (%)	3,80±1,87	3,30±1,49	3,10±1,60	0,594	2-4
Basophils (%)	0±0	0±0	0±0	0	0-1

($p>0.05$) on all hematological parameters, namely hemoglobin, erythrocytes, hematocrit, leukocytes, platelets, count the types of leukocytes (lymphocytes, monocytes, neutrophils, eosinophils), erythrocyte index (MCV, MCH, MCHC), Red Cell Distribution Width (RDW), and blood sedimentation rate (LED) in healthy male volunteers. This is in accordance with the research conducted by Muhamad *et al.*, (2010) where the supplementation of *Pasak Bumi* root showed there were no significant differences in the parameters of hematology (hemoglobin and hematocrit) in healthy male volunteers. Ooi *et al.*, (2015) also reported no significant differences in the types of leukocytes (monocytes, eosinophils, and basophils) after giving supplements of the root of the *Pasak Bumi* to male volunteers.

The results of hematology examination day 0, day 14, and day 42 in healthy women volunteers (Table II) showed no significant differences ($p>0.05$) in hematological parameters, namely hemoglobin, erythrocytes, hematocrit, platelets, count the types of leukocytes (lymphocytes, neutrophils, eosinophils), erythrocyte index (MCV, MCHC), Red Cell Distribution Width (RDW), but showed no significant differences ($p<0.05$) on the parameters of MCH, LED and monocytes. The lack of significant difference in the LED parameters ($p=0.005$) was seen on the 0th day and 14th day of the test, and ($p=0.032$) on the 14th day and 42nd day test, while on the MCH parameter ($p=0.003$) and monocytes (0.009) were seen on the 14th day and 42nd day. The value of the three parameters was still in the normal range.

The examination on day 0 and day 14 to see the effect of the administration of ethanol extracts of *Pasak Bumi* roots capsule for 14 days in healthy female volunteers showed no significant difference ($p>0.05$) on hematological parameters, namely hemoglobin, erythrocytes, hematocrit, leukocytes, platelets, leukocyte count (lymphocytes, monocytes, neutrophils, eosinophils), erythrocyte index (MCH, MCHC) and Red Cell Distribution Width (RDW), and there are no significant differences in the parameters of MCV and LED ($p<0.05$). However, the value of the two parameters is still in the range of normal values.

The result of this study indicates that the administration of ethanol extracts of *Pasak Bumi* root capsules for 14 days has no effect on hematological parameters in healthy male volunteers and healthy female volunteers. This is in accordance with the research conducted by Henkel (2013) where the administration of *Pasak Bumi* root capsules has no effect on hematological parameters in men and women. Hayati (2013) also reported that the administration of capsules of *Pasak Bumi* water extract for 14 days did not affect hematology.

The day 0, day 14, and day 42 examination showed no significant difference in some hematological parameters both in healthy male volunteers and healthy female volunteers, but the hematological values of the hematological parameters were still in the normal range. Hence, it can be concluded that there is no effect on the hematological value before, during, and after consuming capsules of ethanol extracts of

Table II. Mean Values of Hematology of Day 0, Day 14 and Day 42 for Healthy Volunteers for Women (n=10)

Hematology Parameters	Mean Values of Hematology of Healthy Female Volunteers (Mean±SD)			P value	Normal Value
	Day 0	Da 14	Day 42		
Hemoglobin (g/dL)	13,52±0,51	13,43±0,59	13,60±0,48	0.926	11.7-15.5
Erythrocytes (million/uL)	4,82±0,28	4,77±0,36	4,86±0,35	0.836	3.8-5.2
Hematocrit (%)	38,79±1,07	38,7±1,41	39,05±1,41	0.784	35-47
Platelets (10 ⁴ /uL)	28,95±5,55	28,69±6,50	28,95±6,20	0.836	15.0-44.0
MCV (fl)	80,75±5,88	81,49±6,02	80,72±6,06	0.056	80-100
MCH (pg)	28,11±2,09	28,28±2,17	28,10±2,13	0,003*	26-34
MCHC (g/dL)	34,84±0,94	34,69±0,78	34,83±0,65	0.534	32-36
RDW (%)	13,03±0,78	13,12±0,74	13,14±0,73	0.237	11.5-14.5
LED (mm)	13,70±0,78	8,80±4,71	13,50±7,98	0,026*	0-20
Leukocytes (10 ² /uL)	67,88±9,40	73,15±16,17	72,36±22,9	0.385	36.0-110.0
Lymphocytes (%)	35,5±6,96	35,20±9,27	32,40±7,92	0.389	25-4
Monocytes (%)	6,40±1,43	5,60±1,26	6,90±1,37	0,042*	2-8
Neutrophils (%)	54,8±9,24	55,4±11,44	57,60±8,85	0.504	50-70
Eosinophils (%)	3,30±3,13	3,80±3,68	3,10±2,51	0.507	2-4
Basophils (%)	0±0	0±0	0±0	0	0-1

Information: Day 0: Before administering ethanol extract capsules of the *Pasak Bumi* roots; Day 14: After administering ethanol extract capsules of the *Pasak Bumi* roots; Day 42: After 28 days of not administering ethanol extract capsules of the *Pasak Bumi* roots

Pasak Bumi root in healthy volunteers of men and women.

Ismail *et al.*, (2012) reported the total content of protein, glycosaponin, and eurycomanone contained in the roots of *Pasak Bumi* is safe and does not affect the hematological profile (Henkel, 2013).

Monitoring Results of Side Effects of *Pasak Bumi* Root Ethanol Extract Capsules

The clinical symptoms felt by male volunteers for 14 days of taking capsules of ethanol extracts of *Pasak Bumi* are shown in Table III. Clinical symptoms that appear are frequent urination (10%), dizziness (10%), nausea (10%), sweating (10%), insomnia (20%), and increased appetite (20%) of 10 total healthy male volunteers.

The clinical symptoms felt by female volunteers for 14 days of taking capsules of ethanol extracts of the *Pasak Bumi* are shown in Table IV. Clinical symptoms that appear are frequent urination (10%), difficulty sleeping (20%), and difficulty defecating (constipation) (30%) of 10 the total number of healthy female volunteers.

Clinical symptoms that appear in healthy male and female volunteers are in accordance with previous research by George *et al.*, (2016) stating that the use of extracts of *Pasak Bumi* root water at a dose of 200 mg, once a day, for 4 weeks causes

symptoms of insomnia, constipation, and sweating in 40 healthy volunteers. Other studies state that the consumption of ethanol extracts of the root of *Pasak Bumi* in large quantities can cause insomnia, redness of the face, and hyper aggressiveness, but there is no scientific evidence that supports (Li *et al.*, 2013).

On the psychological test during the 14 days of taking capsules of ethanol extracts of the roots of *Pasak Bumi* showed no symptoms affecting the mental condition of male and female volunteers.

Monitoring of side effects continued for 28 days after consuming the last capsule of ethanol extract of *Pasak Bumi* roots. Clinical symptoms experienced by healthy male volunteers on day 14 to day 42 that appear are insomnia (10%), lack of energy (10%), dizziness (10%), and sweating (10%) out of 10 total healthy male volunteers.

Clinical symptoms experienced by healthy female volunteers on day 14 to day 42 that appear are dyspepsia (10%), insomnia (10%), nausea (10%), constipation (20 %), dizzy (20%), and underpowered (20%) of the 10 total number of healthy female volunteers.

The psychological symptom experienced by volunteers, both men and women after 28 days of not taking capsules of ethanol extract of *Pasak Bumi* roots is the feeling of physical fatigue or lack of enthusiasm according to 10% of the total 10

Table III. Frequency of Clinical Symptoms of Healthy Male Volunteers

Clinical Symptoms	Day 0 - Day 14 (n=10)			Day 14 - Day 42 (n=10)		
	Light	Medium	Severe	Light	Medium	Severe
Dyspepsia (Ulcer)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Heat sensation in the body	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Insomnia	1 (10%)	1 (10%)	0 (0%)	1 (10%)	0 (0%)	0 (0%)
Constipation	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Underpowered	0 (0%)	0 (0%)	0 (0%)	1 (10%)	0 (0%)	0 (0%)
Frequent urination	1 (10%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Redness of the face/body	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Dizzy	1 (10%)	0 (0%)	0 (0%)	1 (10%)	0 (0%)	0 (0%)
Nausea	1 (10%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
diarrhea	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Fast heartbeat	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Increased appetite	0 (0%)	2 (20%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Dry mouth	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Allergy	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Mild fever	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Sweating	1 (10%)	0 (0%)	0 (0%)	1 (10%)	0 (0%)	0 (0%)
Total	5 (50%)	3 (30%)	0 (0%)	4 (40%)	0 (0%)	0 (0%)

Table IV. Frequency of Clinical Symptoms of Healthy Female Volunteers

Clinical Symptoms	Day 0 - Day 14 (n=10)			Day 14 - Day 42 (n=10)		
	Light	Medium	Severe	Light	Medium	Severe
Dyspepsia (Ulcer)	0 (0%)	0 (0%)	0 (0%)	1 (10%)	0 (0%)	0 (0%)
Heat sensation in the body	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Insomnia	1 (10%)	1 (10%)	0 (0%)	1 (10%)	0 (0%)	0 (0%)
Constipation	3 (30%)	0 (0%)	0 (0%)	1 (10%)	1 (10%)	0 (0%)
Underpowered	0 (0%)	0 (0%)	0 (0%)	2 (20%)	0 (0%)	0 (0%)
Frequent urination	1 (10%)	0 (0%)	0 (0%)	0 (0%)	1 (10%)	0 (0%)
Redness of the face/body	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Dizzy	0 (0%)	0 (0%)	0 (0%)	1 (10%)	1 (10%)	0 (0%)
Nausea	0 (0%)	0 (0%)	0 (0%)	1 (10%)	0 (0%)	0 (0%)
diarrhea	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Fast heartbeat	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Increased appetite	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Dry mouth	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Allergy	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Mild fever	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Sweating	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Total	5 (50%)	1 (10%)	0 (0%)	7 (70%)	3 (30%)	0 (0%)

Information: Day 0 - Day 14: For 14 days of administering the ethanol extract capsules of Pasak Bumi roots; Day 14 - Day 42: After 28 days of not administering ethanol extract capsules of the *Pasak Bumi* roots

healthy male volunteers and 10 healthy female volunteers. However, this does not significantly affect the physical condition and activities of healthy volunteers.

The long-term administration of capsules of ethanol extracts of *Pasak Bumi* roots in healthy male and female volunteers result in side effects, such as insomnia, frequent urination, constipation,

dizziness, nausea, and sweating, but these side effects occur in a small amount. Long-term use of capsules of ethanol extracts of *Pasak Bumi* roots must be considered and balanced with heavy fluid intake.

CONCLUSION

The administration of capsules of ethanol extract of *Pasak Bumi* roots does not affect the value of hematological parameters, namely hemoglobin, erythrocytes, hematocrit, leukocytes, platelets, count types of leukocytes (lymphocytes, monocytes, neutrophils, eosinophils, basophils), Red Cell Distribution Width (RDW), erythrocyte index (MCV, MCH, MCHC), and blood sedimentation rate (LED) in healthy male and female healthy volunteers. The administration of capsules of ethanol extracts of *Pasak Bumi* roots has side effects in healthy male volunteers in the form of frequent urination, dizziness, nausea, sweating, insomnia, and increased appetite, whereas in healthy female volunteers are frequent urination, insomnia, and constipation.

CONCLUSION

P. niruri ethanol extract gel with added 1% menthol has shown better hair length and weight of the test animals compared to the other gel formulation without menthol, with an average hair length of $12,45 \pm 3,457$ mm and an average hair weight of $28,53 \pm 7,681$ mg, whereas the gel formulation without enhancer yielded $10,67 \pm 2,455$ mm of average hair length and $19,87 \pm 9,552$ mg of hair weight.

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